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## NOTES ON NORTH AMERICAN MYXOSPORIDIA \*

HENRY B. WARD

In this paper are published data on three new species of Myxosporidia. Two species were observed in Lake Erie as parasites of a minnow; the third came from a species of Pacific salmon. Both cases present some unusual features that seem worthy of record. I am greatly indebted to my colleague, Dr. R. Kudo, for valuable assistance given while I was working up the material. The beautiful sketch (Plate V) illustrating the species from Lake Erie was prepared by Mrs. H. S. Jennings, to whom my thanks are due for the courtesy.

### *Myxobolus aureatus* nov. spec. (Plate V)

Host: *Notropis anogenus*.

Location: between the fin membranes.

Locality: near Put-in-Bay, Lake Erie.

Some years ago while engaged in the study of fish parasites for the U. S. Bureau of Fisheries, I discovered a case of infection with a sporozoan parasite which, on examination, proved so unusual in character that careful studies were made of the material then available. The notes made at that time were laid aside in order to secure further specimens and to work out the entire life history. It has proved impracticable as yet to repeat the study of fresh material on the spot and the importance of the find leads me to prepare the data for publication in order that the attention of others may be directed to the species. The form studied departs in some respects from all Myxosporidia yet described and commands attention for certain peculiar biological features.

In August, 1898, while I was seining near the hatchery of the U. S. Bureau of Fisheries at Put-in-Bay, Ohio, some minnows were taken which attracted immediate attention by virtue of their striking appearance. Several species of *Notropis* were netted in the same locality and all were carefully examined. One was conspicuous and only that one was infected in any way. The species in question was

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\* Contributions from the Zoological Laboratory of the University of Illinois, No. 145.



PLATE V

EXPLANATION OF PLATE

*Notropis anogenus* Forbes bearing cysts of *Myxobolus aureatus* in the fins. The cysts in life correspond in color to Japanese gilding. Drawn from life by Mrs. H. S. Jennings, Put-in-Bay, Ohio, August 15, 1898.

determined by Dr. W. C. Kendall, who was serving as ichthyologist of the party, as *Notropis anogenus*, although he noted that these individuals were young and did not agree in all details with the description of that species. These minnows were 2 to 3 centimeters long measured without the caudal fin. In all, thirty specimens of this minnow were captured and seven of these were infected with a myxosporidian parasite. The infected specimens were not inferior in size or vigor to the others of the same species. Even those most severely attacked by the parasite manifested normal activity and responded to experimental stimuli as promptly and accurately as those which showed no sign of being infected. The specimen which was most heavily infected was the most vigorous of all the minnows taken. It lived more than twenty-four hours in a small dish only 4 inches in diameter without any change of water and when killed was still very active.

The infection was markedly conspicuous. At first glance one could see one to many small cysts in the membrane of the fins. They lay between the ectodermal layers of the fin membrane, appearing as brilliant opaque points in the otherwise delicately transparent organ. The cysts were particularly conspicuous because of their striking coloring. Each appeared as an oval body perfectly opaque and glittering like a mass of metallic gold. These cysts were absolutely confined to the fins. Nowhere else on the surface of the body could there be seen even a single such structure and careful dissection failed to disclose any in the flesh elsewhere. Nor were any structures found which could be associated with them even as modified cysts or as developmental stages of the organism. This single stage in the location designated was the only phase in the life history of the organism that I was able to discover. Of the unique character of the location and the color, I shall say more later.

The number of such cysts in the individual case varied widely. In one specimen only a single cyst was present. That was located in the anal fin. In most specimens the cysts were fairly numerous. The individual represented in the plate (Fig. 1) shows the average frequency of infection. It had about thirty-five cysts, distributed as follows: two cysts in the dorsal fin, four in the caudal, eight in the anal, four and two in the two pectoral fins, and three in the single ventral fin present, one of the ventrals being missing in this specimen. The most heavily infected individual had about forty cysts; six of these were located in the dorsal fin, five in the anal, ten in the left pectoral and six in the right pectoral, five in the left ventral and seven in the right ventral. The various specimens showed most distinctly that no uniformity of distribution obtains either with regard to the degree of infection in any particular fin or in respect to the fins infected. Careful

examination of the specimens showed that both the paired and the unpaired fins were infected; the right and left sides proved to be variably infected and any one of the fins might be free from infection although the others were at the same time heavily infected.

In most cases the cysts were clearly separated from each other, though in a few instances they were apparently connected. Even here careful examination of the region under appropriate magnification demonstrated the fact that cysts overlapped in profile only and were actually separate from each other. The cysts were usually single and well separated from those nearest, but in some cases a fin carried a group of two to six cysts rather closely grouped together. There seemed to be no regularity in the occurrence of these groups and as already indicated they were in reality separate cysts though appearing on superficial examination to form a connected mass. As they increased in size the cysts seemed to accumulate chromatophores on the surface. At an early stage when the cyst was small, the chromatophores were few in number; later as the cyst increased in size the chromatophores became much more numerous, and in the largest they were thickly strewn over the surface. Such differences were often seen in adjacent cysts in the same group where the mass of chromatophores imparted a darker, heavier aspect to the older cyst.

The examination of these cysts under a higher magnification showed some interesting structural features. They were exceedingly regular in form and fairly uniform in size although the latter appeared to vary a little with age and development. The individual cyst was a smooth margined ellipsoid, measuring from 1 to 1.6 millimeters in larger diameter and from 0.8 to 1.2 millimeters along its transverse axis (Plate V). The striking color of the living cysts has already been mentioned. Under a high power it seemed to be a clear orange yellow, but under all circumstances was perfectly opaque. The surface of the cyst was spotted with conspicuous black patches of minute size. These spots lay on the outer surface of the cyst wall and were in reality the chromatophores of the skin, but they were distinctly more abundant here than elsewhere in the fin or on the body skin of the minnow. The gilt color was contained in the cyst wall itself, as was easily demonstrated on pulling the structure to pieces. This color faded slowly in alcohol and formol, first losing its brilliancy and later disappearing entirely, leaving the cyst wall a dull white or grayish tone. The cyst wall was noticeably tough and thick in spite of the insignificant size of the cyst. When the wall of the living cyst was torn apart by needles, there exuded a milky white mass from the interior consisting chiefly of the spores to be described later. The gilt color and opacity of the wall remained unchanged.

The presence of color is very unusual in myxosporidian cysts. So far as I can ascertain it is shared by no other species yet described. Recently Southwell and Prashad (1918) reported a cyst of *Myxobolus nodularis* in the muscles of *Rasbora daniconius* as of a creamy yellow color, "in one case appearing blackish owing to the large number of black granules scattered in its surface." These granules are very probably chromatophores on the surface and belong to the host tissue as is indicated in descriptions of other species by various authors; if this inference be correct their record is in part similar to that described here. But the color can hardly be comparable. In fact, as the authors just quoted describe cysts of another species (*Myxobolus rohita*) in the gills of *Labeo rohita* as "of a creamy yellow color," it seems as if they were describing a shade or tone in the preserved specimen rather than a distinct color or pigment; moreover, there is nothing to indicate that the color belongs to the cyst and not to the contents or to the host tissue.

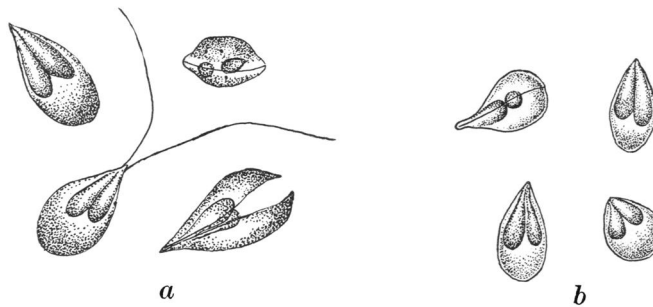


Fig. A.—Spores of *Myxobolus aureatus* drawn from fresh material. *a*,  $\times 1300$ ; *b*,  $\times 972$ .

In discussing such structures various authors agree in stating that the color of the cyst belongs to the host tissues and can be found on examination to disappear when the cyst is removed, thus showing that the true cyst wall is colorless. That is certainly not the condition which obtains in this case, for the color belongs to the cyst and cannot be separated from it in life. In section, the protoplasm shows a poor differentiation into ectoplasm and endoplasm. The former, granular and reticular, covers the entire surface as a thin layer, while the latter is highly vacuolated, containing only mature spores.

When the cyst wall is torn open there exudes a milky white mass composed primarily of the spores. These are characteristic in appearance and demonstrate immediately the myxosporidian nature of the cyst. The spores are ovoid in form (Fig. A), slightly pointed at one end and rounded at the other. The pointed end is the capsular pole. There is no caudal filament present. From one aspect the spore appears

slightly narrower and more pointed than when seen at right angles. Up and down the narrow aspect the spore shows a distinct ridge which marks the line of separation between the two valves of which the spore wall is composed. When the material is left standing in water, the valves separate along this line (Fig. *A*, *a*) and are seen to be perfectly symmetrical and similar in all respects. The shell is of moderate thickness and bears a flange at the lower non-capsular pole. The greatest convexity of the valve is located two-thirds of the way from the pointed pole. The spores vary in length from 12.4 to 13.5 $\mu$  with a breadth of 6.5 to 7.5 $\mu$ , and an average thickness of 5 $\mu$ .

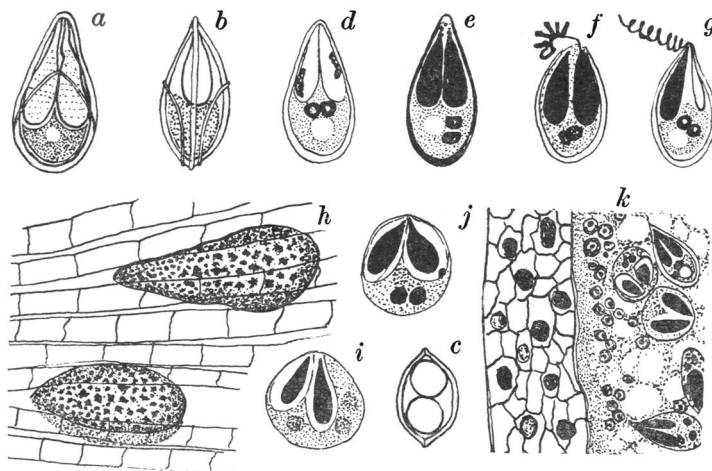


Fig. B.—*Myxobolus aureatus*; magnified 1,500, except as otherwise stated. *a*, *b*, *c*, unstained preserved spores in different views; *d*, *e*, stained mature spores; *f*, *g*, spores with extruded coiled polar filament from section; preparation stained with Giemsa; *h*, a portion of the caudal fin showing two cysts,  $\times 22$ ; *i*, *j*, young spores, stained; *k*, a portion of the cross-section of the fin, showing the peripheral part of the parasite,  $\times 900$ .

Each spore contains two capsules, located in the pointed half of the shell. They are not always exactly alike, for frequently one is slightly longer than the other or else located a little further from the actual pole so that its inner end lies in a different plane from the other. These capsules are elongated pyriform in outline and measure 6 to 7 or rarely 7.5 $\mu$  in greatest length. Ordinarily the filament is not extended, but it can be made to appear by letting the spore stand twenty-four hours or more in plain water. Then one sees two extremely delicate threads, one from each capsule, extending into the water a distance of one and one-half to two times the major diameter of the spore (Fig. *A*, *a*). In the preserved material they may be forced out (Fig. *B*, *f*, *g*) in such fashion as to indicate six or seven coils in the filament. The binucleated

finely granular sporoplasm shows always an iodophilous vacuole which becomes distinctly contoured by taking a deep brownish color when the spore is treated with Lugol's solution. Its diameter is about  $2\mu$ .

The characteristics of this species as described above cause it to fall clearly within the family of the Myxobolidae of Thélohan and the absence of any caudal filament on the spore membrane places it in the genus *Myxobolus* of Bütschli. The polar capsules are equal in size as in the type species *Myxobolus mülleri* Bütschli of Europe which infests many fresh water fishes. Its inclusion in this genus emphasizes its relationship to *M. pfeifferi*, the cause of the devastating barbel disease, and to *M. cyprini*, which gives rise to the destructive fish-pox of the carp.

Only a few forms of the Myxosporidia have been reported within the limits of the United States. Gurley (1893; 111) listed nine species of which eight occur in fresh water hosts. A little earlier Linton (1891) had described a specially interesting form from fresh waters. The species infected was *Notropis megalops* and the locality from which they came was the Black River, Lorain County, Ohio. Since the host is a minnow closely related to that on which occurred the species described in this paper and since the localities are only a short distance apart on the south shore of Lake Erie, one is tempted to ask if the two parasites are not identical. A close examination of Linton's record and figures shows that they cannot possibly be the same species. Linton describes his form as producing globular or botryoidal masses on the side of the head and body and at the base of the fins. The illustration demonstrates clearly the distribution in groups or clusters and further the location of these masses on the body wall at the base of the fins. They occur in the specimen he figured at the base of the pectoral, ventral, anal, dorsal, and caudal fins, but in no case do they encroach on the membrane of the fin itself. They are confined exclusively to the surface of the body proper. The masses are made up of cysts that are distinctly confluent and in no case figured was one cyst discrete and separate from other cysts. The component cysts vary from two to three millimeters in diameter. Finally Linton describes the color of these cysts as white with minute patches of black pigment belonging to the skin of the host.

When these data are compared with those already given for the Put-in-Bay species the differences are conspicuous. The cysts of the latter species are usually single and even when grouped one can distinguish them as separate and entirely unconnected masses. They never form clusters or groups of a botryoidal character. In size they are only



one-third to one-half the dimensions of those in Linton's specimens. In general the cysts described by Linton are much more nearly spherical than those in the present species. Since Linton's material was not living when submitted to him it is uncertain what the appearance was in life, but in the letter from Mr. McCormick of Oberlin College that accompanied the specimens and described their capture no note is made of any color in the living material. It is certainly difficult to believe that any collector could overlook the very brilliant and striking color of the cysts of the Put-in-Bay species so that one may reasonably infer the absence of such coloring. The hosts are different species, one being a river form and the other a lake species, and the lake form was also much smaller than the other for which Linton records a length, exclusive of caudal fin, of 47 to 57 millimeters.

But the most striking difference is found in the location of the cysts. In the Put-in-Bay species they are always in the membranous expansion of the fins and never on the surface of the body, whereas, in Linton's species as already described the location is precisely the contrary. This difference in distribution is uniform and unvarying. No single exception is recorded for either species. The location of the cysts in Linton's species is not uncommon, although most forms that occur on the surface of the body are not confined so rigidly, as his figure indicates this form to be, to the region of the skin just at the base of the various paired and unpaired fins. But the new species described here is found only within the fin membrane, a most unusual location. The significance of this is discussed later, but the marked and constant difference in the location of the cysts may be regarded as clear evidence of the specific difference of the two parasites.

When the spores of the two forms are compared, one finds similar differences. They are, to be sure, much alike in general appearance and structure, but these features are merely those characteristic of all spores in this genus of Myxosporidia. If the drawings of Linton's spores are all of approximately the same magnification as is indicated in the explanation of his plate, then those spores vary in size far more than these. He states the dimensions of the spores in that species as  $17\mu$  long,  $10\mu$  broad, and  $6\mu$  thick, which makes them distinctly larger and different in proportions. They are more drawn out and show a concave taper wanting in the spore from the Put-in-Bay minnow. No comparison can be made of internal structure as he was unable to make out the polar capsules, threads, or nuclei in the spores. In view of all these features it is impossible to include the Put-in-Bay form in the species described by Linton.

The species of *Myxobolus* parasitize the gills, fins, scales, spleen, kidney, and muscles of the host. Commonly they are found in the

connective tissue of these organs and occur in several parts of the body. In our case the very specific localization of the parasite is distinctly noteworthy and in this the species differs from all others in the genus so far as is shown in literature available. The occurrence of Myxosporidian cysts in the fins of fishes is rare indeed. Minchin (1903: 339) cites only five cases: *Henneguya linearis* (Gurley) in *Ameiurus melas* at the base of the dorsal fin; *Myxobolus oviformis* Thél. in the fins, gills, kidney, and spleen of *Gobio gobio*; *Myxobolus mülleri* Bütschli from the fins and gills of *Leuciscus cephalus*; *Glugea acuta* Thél. from the connective tissue of the dorsal fin in *Nerophis aequoreus*, and from the same region in *Syngnathus acus*. From the same genus as our host species Minchin records only one case of infection and that in the skin of *Notropis megalops*, the case described by Linton (1891) and discussed elsewhere in this article. Careful examination of the literature shows that seven cases described as fin infection have been reported up to the present of which, except *Myxobolus seni*, all infect also other organs of the host. These species are as follows:

- Myxobolus sp. Müller (1841: 480)
- Myxobolus oviformis. Thélohan (1895: 351)
- Myxobolus volgensis. Reuss (1906: 200-201)
- Myxobolus gigas. Parisi (1912: 293-294)
- Myxobolus seni. Southwell and Prashad (1918: 347)
- Henneguya linearis var. Gurley (1893: 417)
- Henneguya nüsslini. Schuberg and Schröder (1905: 56)

Of the four cases from the same region in the host, cited after Minchin, the last two concern marine fishes, and the first is doubtful. Of this case, Gurley (1893: 417) speaks as follows in the original description of the species: "In cysts *at the base* of the dorsal fin of *Ameiurus melas* Raf. from Storm Lake, Iowa, a spore occurs which I strongly suspect to be identical with this species, as it answers in every respect to the rather meager diagnosis." As the cyst is *below* and not *in* the fin, the location of the parasite does not at all correspond to that of our species. Gurley also in speaking of *M. linearis* (1893: 416) writes, "Cysts invariably\* embedded in the subcutaneous tissue of some part of the head (especially the under surface of the lower jaw) of *Hybognathus nuchalis* Ag." Here again the location is not that of the species under consideration, as in the former case the cyst is really in the body near the base of the fin. Only one reference in the literature seems to agree in part with the description of *M. aureatus*. Southwell and Prashad (1918: 347) mention that cysts of *Myxobolus seni* were found only "on the median and caudal fins of *Labeo rohita*," a species of fresh water fish taken in Mirpur, India. This is the only species of *Myxobolus* really and exclusively located *in* the fin. Unfortunately

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\*Among several hundred cysts one was seen *at the base* of the pectoral fin.

the authors have given only a very scanty description of this parasite. It is certainly different from the species described here and a detailed comparison is unnecessary.

*Henneguya brachyura* nov. spec.

Host: *Notropis anogenus*.

Location: in the cartilaginous fin ray.

Locality: near Put-in-Bay, Lake Erie.

In studying sections of the caudal fin of one of the minnows that was infected by *Myxobolus aureatus*, a species of *Henneguya* was found encysted in the fin ray. The cysts were rounded with slightly irregular contour and imbedded in the ray. In size they varied from  $160\mu$  in diameter up to  $360$  by  $240\mu$ . No particular cyst membrane could be recognized. The differentiation of the protoplasm into ectoplasm and endoplasm is distinct. The ectoplasm constitutes a layer  $4$  to  $6\mu$  thick, covering the entire surface of the parasite; it shows a

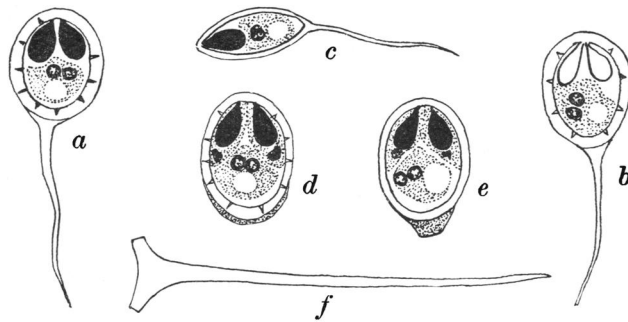


Fig. C.—*Henneguya brachyura*  $\times 1,500$ , except *f*. *a*, *b*, *c*, different views of stained spores from section; *d*, *e*, young spores developing the tail; *f*, detached tail in a section,  $\times 3,560$ .

very finely granular structure. The endoplasm is coarsely alveolar and filled with mature spores in the central portion, while numerous nuclei and young spores in various developmental stages are present in the peripheral portion.

The spore (Fig. C) is a rounded oval in front view but spindle-shaped with symmetrically built valves in profile. The shell is rather thick and the sutural ridge fairly well marked, the sutural edge exhibiting a variable number of folds (8 to 10). The pyriform polar capsules are usually of the same size and form. The tail is a single process, usually more or less bent or irregularly curved, very rarely being straight. In general it is sinuous with two or three shallow curves (Fig. C, *a*) and is rather short, tapering gradually to a point. In young spores which are less deeply stained by any stain, various developmental stages of the tail are easily recognized (Fig. C, *c*, *d*).

Giemsa solution stains the shell proper a clear blue, while the tail takes on a beautiful pink color, showing a distinct difference in affinity for dyes between the material in the tail and in the shell. According to Gurley (1894:250), the tail of *Henneguya macrura*, with which the present species is closely related, was completely dissolved by concentrated sulphuric acid. It seems probable that the tail of this type is entirely different in its development from that of the ordinary bifurcated type, but further studies could not be made in this species owing to the small number of parasites available. In section, dimensions of the species are: length, 10 to 11.5 $\mu$ ; breadth, 8 to 8.75 $\mu$ ; thickness, 4 to 5 $\mu$ ; polar capsules, 3 to 4 by 2 $\mu$ ; length of the tail up to 17 $\mu$ .

Among the known species of *Henneguya*, *H. macrura* Labbé (Gurley, 1894:250) seems to be most closely related to the form under discussion. A comparison of two forms yields the following data:

	<i>H. macrura</i>	The Present Form
Habitat	Subcutaneous connective tissue Head of <i>Hybognathus nuchalis</i> Neches River, Texas, November, 1891	Fin ray of cadual fin; <i>Notropis</i> <i>anogenus</i> ; Put-in-Bay, Ohio, August, 1898
Cyst	Large, elongated; size up to 6 by 2 mm.	Very small; invisible to naked eye; size in section, up to 360 by 240 $\mu$
Spore, similar features	Rounded oval; length 10 to 11 $\mu$ , breadth 6 to 8 $\mu$ , thickness 4 $\mu$ , length of tail 30 to 40 $\mu$	Rounded oval; length 10 to 11.5, breadth 8 to 8.75 $\mu$ ; thickness 4 to 5 $\mu$ ; polar capsules 3 to 4 by 2 $\mu$ ; length of tail up to 17 $\mu$
Differences in the two spores	Sutural ridge without any folds; tail longer, slightly bent; polar capsules larger; valves very un- equal	Sutural edge with distinct folds; tail shorter; sinuous; polar cap- sules smaller; valves usually equal

From this comparison it appears that in form and size the two spores are in close agreement, but the polar capsules differ very distinctly and the valves of the spore are rather sharply contrasted by their nearly equal form in the present type and their unequal form in the older species. Furthermore, the tail of the new form is only half as long as that in *H. macrura* and shows a wavy outline with two or three shallow curves instead of a simple, flat curve as in *H. macrura*. When one adds to these features which distinguish the two spores the radical difference in the size of the cysts, too great to be explained on the basis of differences in age and growth, it is hard to include both in the same species.

Finally Gurley emphasizes the location of the cysts, saying that in *H. macrura* the cysts are "almost invariably situated on some portion of the head," and stating that he had seen "but one exception, a cyst situated at the base of the pectoral fin," whereas the species under consideration was found actually at the opposite end of the body and

only in a cartilaginous ray of the caudal fin. I should not neglect to mention also the difference in hosts and the occurrence of the two parasites in separate geographic provinces. In connection with the morphologic evidence these facts are of significance in contrasting the two forms.

For these reasons I have decided that the new form cannot be brought under the older designation and propose for it the name *Henneguya salminicola*.

*Henneguya salminicola* nov. spec.

Host: *Oncorhynchus kisutch*, the silver salmon.

Location: connective tissue in body muscles.

Locality: taken in Stickeen River, S. E. Alaska.

In connection with studies I am carrying on with the Pacific salmon, the U. S. Bureau of Fisheries sent me preserved specimens labeled

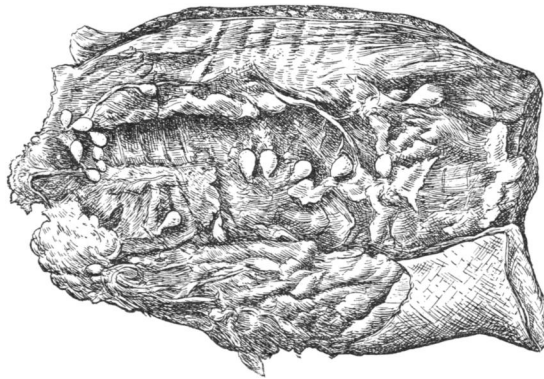


Fig. D.—Cysts of *Henneguya salminicola* in body muscles of Pacific salmon. Approximately half natural size. Preserved specimen.

“Pieces of Salmon with Cysts,” collected by E. Lester Jones, Stickeen River, Alaska, about Sept. 10, 1914. Dr. Jones, who was at that time Deputy Commissioner and engaged in a trip to inspect conditions in Alaskan waters, received the fish within twelve hours of the time they were taken in gill nets so that they were in good condition. The saline solution in which they had been preserved was of a density of 5 or 6 per cent. and had kept the specimens passably well.

On examining the specimen (Fig. D) the observer was at once struck by the pale, whitish flesh around the cysts in clear contrast with the bright pink muscle usual in this fish. The zone of faded tissue surrounded the cysts to a width of 6 to 8 mm. The cysts themselves were pyriform, fairly uniform in size, and hard to the touch. They measured from 3 to 6 mm. in diameter. These cysts were especially conspicuous because some were pendant from the peritoneal wall, and

projected into the body cavity. They are not generally superficial in location as cysts appear everywhere through the muscle mass from the subperitoneal to the subdermal connective tissue, though of course all are subperitoneal in position. While they occur in groups in a certain sense, each cyst is entirely independent of those near it so far as can be determined by the unaided eye or by dissection. They certainly do not form botryoidal masses such as are found in some cases.

Sections demonstrate that the cysts are surrounded by a heavy capsule of connective tissue. Spores in various stages of development occur within the capsule and the mature spores are thickly massed together in the central area of the cyst.

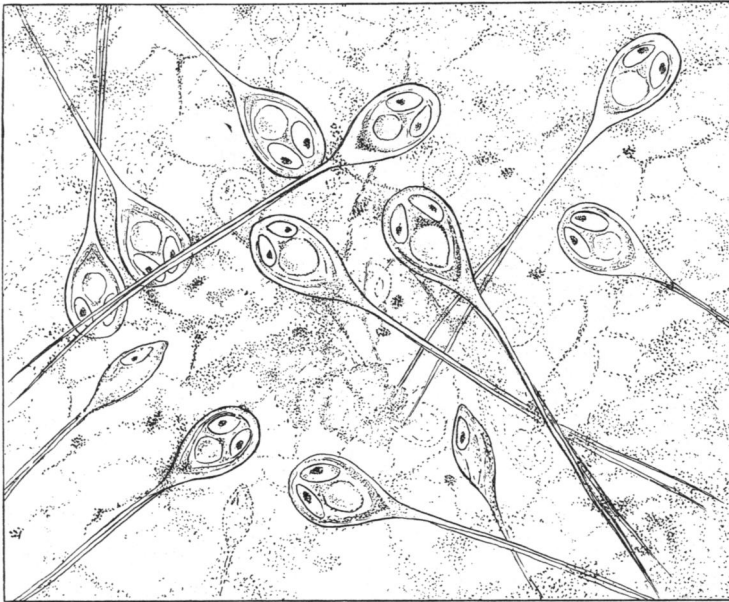


Fig. E.—Spores of *Henneguya salminicola* from section of cyst stained with iron hematoxylin.  $\times 1,180$ .

There was, of course, no chance to study living material. The form of the spores is clearly shown in a drawing (Fig. E) made from a section of the cyst contents; the slide had been stained in iron hematoxylin and picric acid. The two long and delicate spines that project from the non-capsular end of the spore are in reality prolongations of the shell that are not pierced by the cavity of the spore. This form is characteristic of the genus *Henneguya*. Here the caudal spines are separate throughout their entire length but are roughly parallel and not divergent. The two nearly equal polar capsules are not contiguous along the median line but are separated by a band one-third to one-half the width of a capsule. A large iodophilous vacuole, 3.4 to 4  $\mu$  in

diameter, is conspicuous in the spore, but the polar filament coiled in the capsules cannot be distinctly seen in the preserved material.

A series of careful measurements was made of the spores and their processes. The body of the spore when measured in stained specimens "over all" varied in length from 11.97 to 14.25 $\mu$ , on the average being 12.42 $\mu$ , though the norm of length as calculated from the series was very close to 12 $\mu$ . If measured to the base inside, stained specimens are 8.4 to 8.66 $\mu$ . The width of the body of the spore varied from 7.12 to 8.43 $\mu$ , with an average of 7.92 $\mu$  and a norm of 8 $\mu$ . The length of the tail was from 30.78 to 38.19 $\mu$ , with an average of 34.54 $\mu$  and a

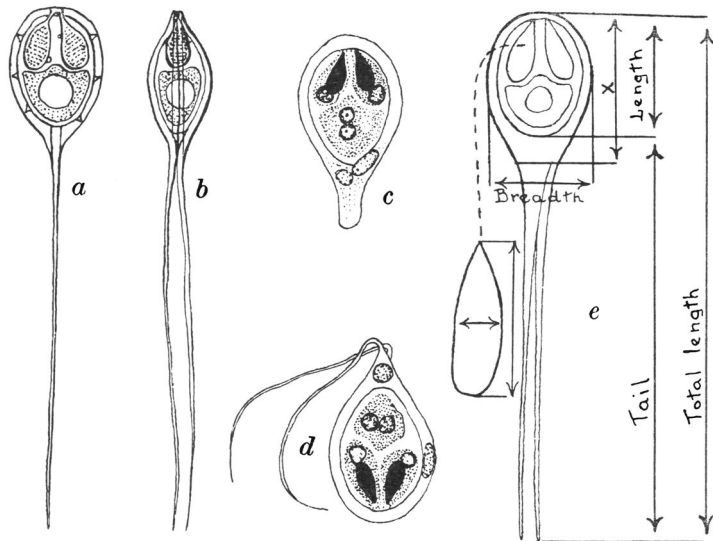


Fig. F.—*Hennequya salminicola*; a, b, unstained preserved spores; c, young spore; d, more advanced stage from smear stained with Giemsa; a-d,  $\times 1,500$ ; e, diagram showing limits observed in taking measurements, x, the length "over all," is not often used since the lower limit is not definitely marked.

norm of 35 $\mu$ . In another set of thirteen specimens the average length was 12.44 $\mu$ , the length of the tail 35.49 $\mu$ ; the average width of seven specimens was 7.63 $\mu$  and the thickness of six specimens averaged 4.78 $\mu$ .

The polar capsules range from 3.70 to 4.56 $\mu$  in length by 1.59 to 2.85 $\mu$  in breadth, or in the norm 4 by 1.8 $\mu$ . One is almost always a little larger than the other, the difference being constantly about 0.25 $\mu$  in length and half as much in width. Very few exceptions to this rule were met in a long series of measurements.

Since differences actually are found between measurements of spores of the same species in stained and unstained preparations, I give a table showing results obtained by two observers with different technic. The diagram (Fig. F, g) shows the limits used in making the measure-

ments recorded. A series of twenty-four to thirty mature spores was used in each case.

MEASUREMENTS OF SPORES OF *HENNEGUYA SALMINICOLA*

Measured by .....	Camera drawing	Ocular	Micrometer
Stained by .....	Iron hematoxylin	Giemsa	Unstained
Total length .....	42.75-52.44	43.5 -53.0	51 - 57
Length of } "Over all" .....	11.97-14.75	.....	.....
spore body } Inside base .....	.....	8.4 - 8.66	8.6 - 9.5
Length of tail .....	30.78-38.19	34.25-36.75	.....
Breadth of spore body .....	7.12- 8.43	7.5 - 8.5	8.6 - 9.5
Polar } Length .....	3.7 - 4.56	3.5 - 3.8	3.5 - 4.0
capsule } Breadth .....	1.59- 2.85	1.7 - 2.2	2.0 - 2.5

All measurements expressed in microns.

In form and size of spores this species resembles most closely *Henneguya zschokkei*, *H. schizura*, and *H. nüsslini*. *Henneguya nüsslini* was discovered in the trout by Schuberg and Schröder (1905), from whose description the following data are excerpted. The two cysts found lay in the subcutaneous connective tissue at the base of the dorsal fin. The spores were  $12\mu$  long by 8 to  $9\mu$  broad, and with the tail measured  $32\mu$  over all. The tail was split, but the two spines were never separated throughout their entire length. The polar capsules were  $5\mu$  long and  $3\mu$  wide; they do not meet along the median line, but are separated by a distinct space. The spore is rounded at the anterior end. In this respect and in the separation of the polar capsules the new species is like *H. nüsslini* and unlike the other species named above, but a comparison of the dimensions quoted shows that *H. nüsslini* has larger polar capsules and a larger spore body, whereas the total length is much less than in *H. salminicola*. These differences are too great to permit including the new form in the species *H. nüsslini*.

*Henneguya schizura* was first described by Johannes Müller but was not named until Gurley (1893:417) called it *Myxobolus schizurus*. The parasite is found only in the orbit, encysted in the connective tissue of the eye muscles, in the sclerotic and between the latter and the choroid. It occurs in young *Esox lucius* and is present in May and June. Müller looked for it without success in specimens of the pike from North America. The two species agree in the length of the spore body ( $12\mu$ ), but in *H. schizura* the spore is only  $6\mu$  broad as against  $8\mu$  here, and the tail in the former is three to four times as long as the spore body, whereas here it is barely three times as long. The size of the spores is sufficient in fact to distinguish this form from the new species described here although the peculiar and restricted distribution of *H. schizura*, occurring only in the orbital tissue, precludes the possibility of considering the new species identical with it.

A close resemblance exists between the new species and *Henneguya zschokkei* which was first described by Zschokke and named by Gurley



(1893). It inhabits the subcutaneous and superficial connective tissue in the trunk muscles of *Coregonus fera*. The cysts are round or oval and of considerable size, up to 30 mm. in maximum. The spore body is  $10\mu$  long by  $7\mu$  broad. The tail is four to five times as long as the spore body and is composed of two slightly curved and diverging spines. In comparison with the species from the Pacific salmon, *H. zschokkei* has a smaller spore body and a longer tail. In the former species the tail filaments are nearly parallel and never divergent as in *H. zschokkei*. These points form an adequate basis for separating the two forms.

It is worthy of note that such parasites though common in many types of fish are almost entirely unknown in salmon of any sort. An examination of the literature shows only a single record of a Myxosporidian parasite in any European salmon. That is *Lentospora cerebralis* which is the cause of the gid disease (Drehkrankheit) of young salmon in the first year of life.

I have not been able to find a single record of the occurrence of Myxosporidia in an adult European salmon and not one in a salmon of any age from this continent. During the last fifteen years I have examined personally several thousand Pacific salmon of all species and have never seen one infected so far as could be detected with the unaided eye. In searching for diseased fish I have been aided by a large number of fishermen and other cannery employees who knew I was anxious to secure all such specimens and were desirous of aiding me so that the cases I have recorded represent those culled from several hundred thousand fish, and there is no entry in my notes of a Pacific salmon affected with any sort of myxosporidian disease.

It is hardly possible that this case could represent a seasonal disease which fell outside the time limits of my experience, for I have collected salmon in the Alaskan coastal waters at least as late as September 1, and the date of this find was only ten days later. Further, no report of such a condition has been transmitted to me by the many men in that region who have been interested in my work and anxious to participate in it.

Finally, one must consider the chance that this is a localized disease and infects only or chiefly the salmon that run in the Stickeen River. I have not collected or studied the Pacific salmon in that precise region and so cannot venture to pass judgment on the question. But if the infection is localized it must be held within narrower limits than are usually observed by the parasites of migratory or marine fish so far as I know them; for I have studied the salmon run both north and south of the Stickeen River and the channels connecting with it, and the fish boats which supplied the canneries at which I was working ranged nearly as far as the Stickeen; yet no fish were seen with a similar infection.

So far as I can ascertain, this is the first published record of the occurrence of a myxosporidian parasite in any fish from Alaskan waters. While the lack of records is very likely due in part to the limited attention paid to diseases of fish from that region, I am also inclined to believe, from my own observation, that myxosporidian parasites are rare in fish found in Alaskan coastal waters.

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